

POSSIBILITY OF USING THE BOTANICAL INSECTICIDE AZADIRACHTIN
AND SYNTHETIC AND SEMI-SYNTHETIC INSECTICIDES TO CONTROL
HELICOVERPA ARMIGERA IN SWEET PEPPER

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Abstract: During 2019 and 2020, field experiments were performed on sweet pepper crops to determine the efficacy of chemical insecticides (lambda-cyhalothrin, flubendiamide), semi-synthetic (emamectin benzoate), and biological pesticide (azadirachtin) in controlling cotton bollworm (*Helicoverpa armigera*). The experiments were performed in a randomized complete block design with four replications according to the standard EPP0 method at the site of Veliko Gradište (Serbia). Flubendiamide was applied at a rate of 50 g/ha, lambda-cyhalothrin at a rate of 7.5 g/ha, emamectin benzoate at a rate of 375 g/ha, and azadirachtin at a rate of 0.75 g/ha. The intensity of the 2nd generation cotton bollworm infestation on sweet pepper at this locality was higher during 2020 compared to 2019. After performing two treatments for the 2nd generation, flubendiamide showed the highest efficacy, ranging from 92.42% (3 days after treatment – DAT, 2020) to 95.56% (9DAT, 2019). Lambda-cyhalothrin had a satisfactory efficacy in the range of 81.93% (9DAT, 2020) to 90.63% (3DAT, 2019), and emamectin benzoate showed similar efficacy from 80.72% (9DAT, 2020) to 90.63% (3DAT, 2019). Azadirachtin could gain a significant place as a botanical insecticide in integrated pest management programs for sweet pepper protection from *H. armigera*. However, it statistically showed a significantly lower efficacy than other insecticides (77.27%: 3DAT, 2020 to 86.67%: 9DAT, 2019).

Key words: effects, insecticides, botanical insecticide, cotton bollworm, *Capsicum annuum*.

Introduction

The cotton bollworm (*Helicoverpa armigera* Hübner, Lepidoptera: Noctuidae) is a polyphagous pest (Fathipour and Sedaratian, 2013) with high fecundity, a wide range of host plants, and a high potential for developing insecticide resistance. The larvae cause damage to peppers by burrowing into fruits and feeding on their

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internal contents. Fruits damaged in this way are susceptible to attack by pathogens that cause wilting and rot (*Fusarium* spp., *Alternaria* spp., *Erwinia carotovora*). As such, they are not suitable for human consumption (Sekulić et al., 2004). The producers must pay special attention to cotton bollworm monitoring and control as it can potentially cause significant damage. Various non-chemical (agrotechnical, mechanical, biological) and chemical measures are used to control *H. armigera*. Of all the available measures, the application of insecticides is the most common, especially in the conditions of growing sweet peppers in the field. However, for insecticides to give good results, treatments must be timely, i.e., they should be performed when earlier larval stages are present and before they penetrate the fruits.

Various insecticides are available worldwide for the control of *H. armigera*. These include non-selective pyrethroids and newer, highly selective diamides, semi-synthetic insecticides, and some compounds of natural origin. Natural products have been increasingly used to control various pests of cultivated plants. Bioinsecticides have some favorable characteristics such as a biological basis and an excellent toxicological and ecotoxicological profile. Azadirachtin belongs to the biochemical bioinsecticides and is one of the limonoids extracted from the Indian neem tree, *Azadirachta indica*. Structurally, azadirachtin is similar to the insect hormone ecdysone and labeled as an ecdysone blocker. The antifeeding effect occurs because of the action of azadirachtin on the taste receptors and the paralysis of the oral apparatus, which results in the termination of feeding (Mordue et al., 2010). Azadirachtin is widely used in the world for the protection of various cultivated plants because of its biological origin, its favorable toxicological and ecotoxicological properties, different modes of action, low potential for pest resistance development, and promising effectiveness in controlling pests from distant insect orders (Lepidoptera, Coleoptera, Hemiptera, and Homoptera). Flubendiamide and emamectin benzoate, semi-synthetic bioinsecticide, are highly selective insecticides with exceptional biological activity in controlling Lepidoptera larvae, especially *H. armigera* (Ameta and Bunker, 2007). In addition, their selectivity for beneficial arthropods makes them suitable for inclusion in integrated pest management (IPM) programs. This contrasts with pyrethroids, which damage beneficial arthropods, and where resistance problems develop. *H. armigera* is a species whose populations rapidly develop resistance to insecticides. This pest has developed resistance to many synthesized chemicals from the group of pyrethroids, organophosphates, carbamates, etc. The resistance of the cotton bollworm to pyrethroids was found in southern France, where the resistance factor for deltamethrin was 32-fold (Buès et al., 2005). High resistance to cypermethrin was also found in southern India, where the resistance factor for this compound was 48-fold, while resistance to chlorpyrifos was low to moderate (Chaturved, 2007). The ecotoxicological and toxicological consequences of resistance manifest through environmental pollution due to the increase in insecticide application rates

and the increased number of treatments during the growing season. These consequences are critical reasons for the mandatory introduction of natural products in IPM plant protection programs, where bioinsecticides and semi-synthetic products are crucial.

This study aimed to determine the efficacy of insecticides of chemical, semi-synthetic and biological origin in the control of cotton bollworm (*H. armigera*) on sweet pepper in field conditions to justify their use.

Material and Methods

The field trials were performed according to the standardized and partially adapted EPPO method PP 1/295 (1) (EPPO Standards, 2016) to test insecticide efficacy in controlling *H. armigera* on vegetables and ornamentals (EPPO).

The experiments were carried out during 2019 and 2020 in the sweet pepper crops at the locality of Veliko Gradište (GPS: N 44° 44.477204; E 21° 24.177048) in a randomized complete block design with four replications. The size of the experimental plot was 25 m². The “Solo” backpack sprayer was used for the treatments. The insecticide preparations were applied with a water consumption of 500 l/ha. The effects of the following insecticides were investigated: flubendiamide, emamectin benzoate, lambda-cyhalothrin, and azadirachtin (Table 1).

Table 1. Insecticides examined in the trials.

Insecticide	Trade names of the insecticides (the content of a.i.)	Amount of insecticide preparation	Amount of insecticide (g/ha)
Flubendiamide	FLUBENDIAMIDE SC (200 g a.i./l)	0.25 l/ha	50
Emamectin benzoate	AFFIRM 095 SG (250 g a.i./l)	1.50 kg/ha	375
Lambda-cyhalothrin	LAMDEX 5 CS (50 g a.i./l)	0.15 l/ha	7.5
Azadirachtin	NIMBECIDINE EC (0.3 g a.i./l)	2.5 l/ha	0.75
Control (untreated)	-	-	-

Two treatments of the sweet pepper crop were performed to control the second generation of cotton bollworm during each year of examination. The first treatment was established based on monitoring the flight of the butterflies with light traps and visual inspection of the fruit to determine the presence of pest eggs. The first treatments were performed after the confirmed presence of laid eggs, and the first hatched larvae before their penetration into the fruits. The treatments were carried out in the evening to ensure optimal temperature conditions for the insecticides to take effect and to avoid direct sunlight. The usual agrotechnical and plant

protection measures were implemented since the crop was established. The first generation of *H. armigera* was regularly controlled. The evaluation of the trial results was performed in two periods: three days after the second treatment (3DAT) and nine days after the second treatment (9DAT) (Table 2).

Table 2. Dates of insecticide treatments and result evaluations.

Locality	Veliko Gradište, Serbia	
Year	2019	2020
Date of the first treatment	August 6	August 12
Date of the second treatment	August 15	August 21
The first evaluation	August 18 (3DAT*)	August 24 (3DAT)
The second evaluation	August 24 (9DAT)	August 30 (9DAT)

*DAT: days after the second treatment.

The evaluation was conducted by examining 100 randomly selected fruits in each experimental plot and determining the number of fruits damaged by *H. armigera* larvae.

The standard EPPO method PP 1/152 (4) (EPPO Standards, 2012) was used for the statistical processing of the test results. The average damage of fruits (M_s) by treatments and the standard deviation (S_d) were determined, as well as the comparison of means, i.e., the significance of the differences between the treatments (Student's t-test). The analysis of variance was processed in the Microsoft Excel computer program. The percentages of fruit damage in treatment replications (x) were previously transformed using statistics: $\sqrt{x + 0,5}$.

Immediately before each treatment, it was found that there were no damaged fruits, only a certain number of eggs laid. Therefore, the efficacy of the insecticides was calculated using the Abbott's formula based on the damage observed in the post-treatment assessments.

Results and Discussion

The test results are shown in Tables 3 and 4.

At the locality of Veliko Gradište, the intensity of infestation of the second generation of the cotton bollworm on sweet pepper was higher during 2020 compared to 2019, so that the average fruit damage in the untreated plot was 8% (3DAT) and 11.25% (9DAT) (Table 3), while in 2020, it was 16.5% (3DAT) and 20.75% (9DAT) (Table 4).

In our experiments, lambda-cyhalothrin (7.5 g/ha) had a satisfactory efficacy in the control of *H. armigera* on sweet pepper at the locality of Veliko Gradište,

but the efficacy was better at lower levels of this pest. In the first assessment after the second treatment (3DAT), the efficacy of this compound was 90.63% (2019) and 86.36% (2020). In the later assessment (9DAT), there was a particular decline in the degree of efficacy, and it amounted to 88.89% (2019) and 81.93% (2020). The field trials conducted during 2012 and 2013 confirmed that the efficacy of lambda-cyhalothrin ranged between 88.03% and 90.89% in controlling the cotton bollworm on chickpea (Yogeeswarudu and Venkata Krishna, 2014).

Table 3. The efficacy of the insecticides applied in the control of *H. armigera* on sweet pepper (Veliko Gradište, 2019).

Treatments	Average damage of fruits (Ms ± Sd)*	
	<i>Efficacy %</i>	
	3DAT	9DAT
Lambda-cyhalothrin (7.5 g/ha)	0.75 ^{a**} ± 0.96 90.63	1.25 ^a ± 0.96 88.89
Flubendiamide (50 g/ha)	0.5 ^a ± 0.58 93.75	0.5 ^b ± 0.58 95.56
Emamectin benzoate (375 g/ha)	0.75 ^a ± 0.96 90.63	2.0 ^c ± 0.82 82.22
Azadirachtin (0.75 g/ha)	1.25 ^b ± 0.96 84.38	1.5 ^b ± 0.58 86.67
Control (untreated plot)	8.0 ^c ± 1.83	11.25 ^d ± 3.59
LSD _{0.05}	0.1182	0.1033
LSD _{0.01}	0.1957	0.1709

*Data are expressed as average damage of fruits (Ms) ± standard deviation (Sd) of four replications of each insecticide treatment; **Mean values followed by the same superscript letter (s) within the same column are insignificantly different ($P \leq 0.05$; $P \leq 0.01$) according to the Student's *t*-test.

Of all the tested insecticides, flubendiamide (50 g/ha) showed the highest efficacy during both years. In the evaluations at 3DAT and 9DAT, flubendiamide showed good efficacy, and it amounted to 93.75% and 95.56% during 2019. The excellent efficacy of this insecticide was also recorded in 2020, namely 3DAT: 92.42% and 9DAT: 93.98%.

During 2019 and 2020, emamectin benzoate (375 g/ha) showed a statistically significantly lower efficacy than flubendiamide in our experiments, while it also had a weaker prolonged effect at 9DAT. It had a satisfactory initial efficacy (3DAT) of 90.63% (2019) and 87.88% (2020). The weaker efficacy of emamectin benzoate was achieved at 9DAT, and it amounted to 82.22% (2019) and 80.72% (2020).

According to the results of the field trials conducted during 2005 and 2006 in the state of Tamil Nadu (India), the efficacy of flubendiamide (Flubendiamide 480 SC, 125 ml/ha) in controlling *H. armigera* on tomatoes ranged from 86.24% to

99.51%, while emamectin benzoate had a lower efficacy (64.14–79.76%) (Kubendran et al., 2008). Ameta and Kumar (2008) reported the excellent efficacy of flubendiamide in controlling the cotton bollworm on the chili pepper. Murugaraj et al. (2006) emphasized a high efficacy of 91.46% for emamectin benzoate in the control of *H. armigera* on tomatoes.

Table 4. The efficacy of the insecticides applied in the control of *H. armigera* on sweet pepper (Veliko Gradište, 2020).

Treatments	Average damage of fruits (Ms ± Sd)	
	<i>Efficacy %</i>	
	3DAT	9DAT
Lambda-cyhalothrin (7.5 g/ha)	2.25 ^a ± 0.96 86.36	3.75 ^a ± 1.50 81.93
Flubendiamide (50 g/ha)	1.25 ^b ± 0.50 92.42	1.25 ^b ± 1.26 93.98
Emamectin benzoate (375 g/ha)	2.0 ^c ± 0.82 87.88	4.0 ^c ± 0.82 80.72
Azadirachtin (0.75 g/ha)	3.75 ^d ± 0.50 77.27	4.25 ^d ± 0.96 79.52
Control (untreated plot)	16.5 ^e ± 4.20	20.75 ^e ± 3.30
LSD _{0.05}	0.0393	0.0288
LSD _{0.01}	0.0651	0.0477

In our experiments during 2019 and 2020, azadirachtin (0.75 g/ha) showed a statistically significantly lower efficacy compared to the other tested insecticides ($P \leq 0.05$; $P \leq 0.01$), with a more pronounced and prolonged effect at 9DAT. However, although its efficacy was satisfactory or even weak in the conditions of less persistent attacks of *H. armigera*, it offers certain advantages over the synthetic insecticides due to its biological origin. Azadirachtin showed an efficiency of 84.38% (2019) and 77.27% (2020) at the 3DAT assessments. In the later assessment at 9DAT, there was an increase in the degree of efficacy, and it amounted to 86.67% (2019) and 79.52% (2020). In trials conducted by other authors during 2013 and 2014, the efficacy of azadirachtin A (Nimbecidine, 0.4%) in the control of *H. armigera* on tomatoes was 83.33% (Kumar et al., 2016). Yankova and Todorova (2011) found that the efficacy of azadirachtin (NeemAzal T/S 0.3%) was 77.12% in controlling the cotton bollworm on peppers. Gayi et al. (2016) discussed the excellent efficacy of azadirachtin and bifenthrin in *H. armigera* larvae control on cotton. Good efficacy of azadirachtin and emamectin benzoate in the control of *H. armigera* on tomatoes was also found by Shah et al. (2013). An efficacy of 87.37% in the control of *Tuta absoluta* was achieved by applying azadirachtin in tomatoes (Sammour et al., 2018).

Conclusion

All tested insecticides could be found within the regular IPM programs to protect sweet pepper from the cotton bollworm, depending on the intensity of the infestation by this pest. Flubendiamide as the most effective tested insecticide with a short withdrawal period (3 days) would be a suitable solution in conditions of intensive activity of *H. armigera*, especially the second generation, but also later generations that occur when the harvest has already begun. Lambda-cyhalothrin exhibited poorer efficacy in intense cotton bollworm attack conditions, so it could be positioned in IPM programs when the pest population density is lower, and this is mostly the typical case during the development of the first generation. The semi-synthetic insecticide emamectin benzoate and the bioinsecticide azadirachtin have shown lower efficacy in severe infestation of *H. armigera*. However, they should also be included in IPM programs due to their favourable ecotoxicological and toxicological properties. They could be recommended for sweet pepper protection when this pest is present in lower population densities. In addition, due to the short withdrawal period, biological insecticides should be used in case of pest activity at the beginning of ripening and between fruit harvests.

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MOGUĆNOST PRIMENE BIOINSEKTICIDA AZADIRAHTINA I NEKIH
SINTETIČKIH I POLUSINTETIČKIH INSEKTICIDA U SUZBIJANJU
HELICOVERPA ARMIGERA NA PAPRICI

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R e z i m e

Tokom 2019. i 2020. godine vršeni su poljski ogledi na usevima paprike u cilju utvrđivanja efikasnosti sintetičkih insekticida (lambda-cihalotrin, flubendiamid), polusintetičkih (emamektin benzoat) i bioinsekticida (azadirachtin) u suzbijanju pamukove sovice (*Helicoverpa armigera*). Ogledi su izvedeni prema tipu potpunog slučajnog blok sistema u četiri ponavljanja prema standardnoj metodi EPPO na lokalitetu Veliko Gradište (Srbija). Flubendiamid je primenjen u količini od 50 g/ha, lambda-cihalotrin u 7,5 g/ha, emamektin benzoat u 375 g/ha, a azadirachtin u količini od 0,75 g/ha. Intenzitet infestacije larvama druge generacije pamukove sovice na paprici na ovom lokalitetu bio je veći tokom 2020. godine u odnosu na 2019. godinu. Nakon obavljena dva tretiranja za suzbijanje druge generacije, flubendiamid je pokazao najveću efikasnost, u rasponu od 92,42% (3 dana posle tretiranja – DPT, 2020) do 95,6% (9DPT, 2019). Lambda-cihalotrin je imao zadovoljavajuću efikasnost u rasponu od 81,93% (9DPT, 2020) do 90,63% (3DPT, 2019), a emamektin benzoat je pokazao sličnu efikasnost od 80,72% (9DPT, 2020) do 90,63% (3DPT, 2019). Azadirachtin bi kao bioinsekticid mogao da zauzme značajno mesto u integralnim programima zaštite paprike od *H. armigera*. Međutim, statistički je pokazao značajno nižu efikasnost od drugih insekticida (77,27%: 3DPT, 2020, do 86,67%: 9DPT, 2019).

Ključne reči: efekti, insekticidi, bioinsekticid, pamukova soвица, *Capsicum annuum*.

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